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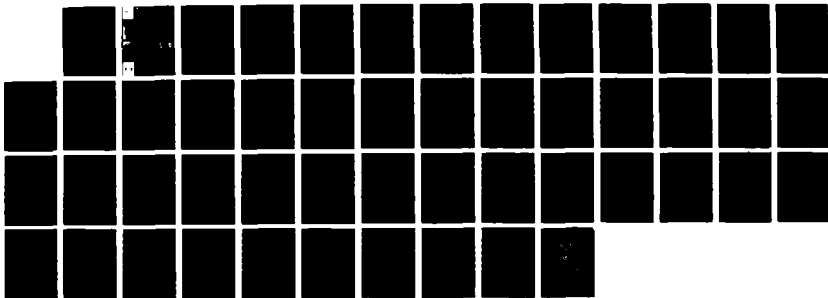
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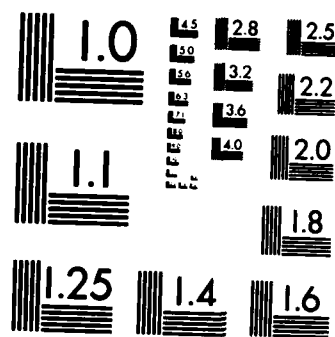
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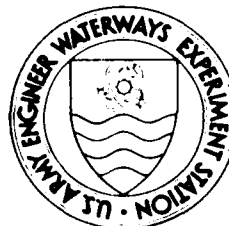
THE USE OF FERTILIZER TO ENHANCE TRANSPLANTS OF THE SEAGRASSES *ZOSTERA MARINA* AND *HALODULE WRIGHTII*

by

Mark S. Fonseca, W. Judson Kenworthy,
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Beaufort, North Carolina 28516

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<p>A recent need to restore seagrass systems has resulted in numerous attempts to transplant most of the North American seagrass species. Transplanting technology also has received increased attention, but the potential of increasing coverage through fertilizer subsidies has not been adequately addressed. This study examined the influence of slow-release fertilizers (14-14-14, balanced, and 18-0-0, unbalanced--percentage nitrogen, phosphorus, and potassium, respectively) on survival, population growth, and areal coverage of the seagrasses <i>Zostera marina</i> and <i>Halodule wrightii</i> as well as flowering and productivity of some <i>Zostera</i> plantings.</p> <p>Three experimental plantings were conducted on each of three sites near Beaufort, N. C. Plantings were done in fall 1984 and spring 1985 with <i>Zostera</i> and in late spring 1985 with <i>Halodule</i>. The two fertilizer types were applied in three dose levels (10, 90,</p> <p style="text-align: right;">(Continued)</p>				
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and 170 g/planting unit). Replicate treatments, including fertilized control plantings, were randomly assigned locations on each of the three sites. Release of fertilizer, as well as light energy, temperature, salinity, and sediment stability, was monitored during the course of the study.

No pattern of population growth or coverage could be found that could be ascribed to differences in environmental conditions. Nitrogen was released roughly according to the manufacturer's specifications for the 14-14-14 applications. Release of the 18-0-0 did not occur in the spring *Zostera* plantings, apparently due to quality control problems with its manufacturing. Phosphorus did not release in any of the experiments. A significant difference was found in population growth between pooled balanced and pooled unbalanced fertilizer treatments in the fall *Zostera* experiment only. Some differences were found between fertilizer treatments for areal coverage as well. Percent flowering of *Zostera* did not differ as a function of fertilization, but spring plantings were significantly greater than fall plantings. This suggested that fall plantings would suffer less from flowering-induced mortality. Productivity of balanced fertilizer treatments was significantly greater than unbalanced and control treatments as examined in the fall *Zostera* planting.

The lack of phosphorus release meant that nitrogen release would have to account for the observed differences in population growth, areal coverage, and productivity observed between fertilizer treatments. Since the quantity of nitrogen release overlapped significantly between the two fertilizer types, a logical explanation for the observed differences in the seagrass growth responses could not be provided. The lack of phosphorus release also casts doubts on the validity of previous studies using similar fertilizer types where the fertilizer composition and release were not examined over time. It was concluded that no alterations in plant spacing should be considered and that, until further studies are conducted, management and restoration plans should consider fertilizer subsidies only as experimental pilot studies.

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PREFACE

This report was sponsored by the Office, Chief of Engineers (OCE), US Army, as a part of the Environmental Impact Research Program (EIRP), Work Unit entitled Coastal Erosion Control Techniques Using Plants. The Technical Monitors for the study were Dr. John Bushman and Mr. Earl Eiker of OCE and Mr. David B. Mathis, Water Resources Support Center.

The study and preparation of a draft final report were accomplished during the time period 1 October 1984 and 1 October 1985; preparation of the reproducible copy was done during February 1986.

The report was prepared by Mark S. Fonseca, W. Judson Kenworthy, Keith Rittmaster, and Gordon W. Thayer of the Southeast Fisheries Center, Beaufort Laboratory, Division of Estuarine and Coastal Ecology, National Marine Fisheries Service, under Support Agreement WESCW 85-60.

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Dr. Thomas J. Fredette, Coastal Ecology Group, was the WES contract monitor for the research, under the general supervision of Mr. E.J. Pullen, Chief, Coastal Ecology Group, and Dr. C.J. Kirby, Chief, Environmental Research Division. Dr. Roger T. Saucier, WES, was the Program Manager of EIRP.

COL Allen F. Grum, USA, was the previous Director of WES. COL Dwayne G. Lee, CE, is the present Commander and Director. Dr. Robert W. Whalin is Technical Director.

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THE USE OF FERTILIZER TO ENHANCE TRANSPLANTS OF THE SEAGRASSES
ZOSTERA MARINA AND HALODULE WRIGHTII

PART I: INTRODUCTION

1. A recent surge in mitigation interest and need to restore seagrass systems has resulted in numerous efforts to transplant Zostera marina and Halodule wrightii. The need to transplant these and other seagrasses has emerged from coastal development pressures that have either removed or deteriorated the seagrass environment (Thayer et al. 1975; 1984; and in press). Present logistics of transplanting technology have been examined extensively (Fonseca et al. 1982; 1984; 1985; in press), but the potential of increasing coverage through nutritional subsidies once transplants have been established has not been adequately addressed. The manner in which seagrasses should be subsidized, however, must be considered in view of the relatively limited (in comparison to terrestrial crops) knowledge of their growth requirements.

2. The target species of this study, eelgrass (Zostera marina) and shoalgrass (Halodule wrightii), form highly productive communities that occur in shallow waters of North America (Thayer et al. 1984). Zostera dominates the temperate seagrass community on both the Atlantic and Pacific coasts, while Halodule occurs in the United States, throughout Florida, the Gulf Coast, and the U.S. Virgin Islands. These two species co-occur only along a narrow band of the middle Atlantic shoreline extending from Ocracoke Inlet, N. C., south to Cape Fear, N. C. (Thayer et al. 1984). Like most other seagrasses, these two species grow rooted in unconsolidated sediments. Zostera and perhaps most of the other seagrasses have a well-developed and functional vascular system that enables them to transport gases and nutrients internally (Penhale and Wetzel 1983, Pregnall et al. 1984, Smith et al. 1984). Nutrients are absorbed by either the roots or leaves (McRoy et al. 1972, Short and McRoy 1984, Thursby and Harlin 1982); hence, these plants are able to derive nutrition from both the water column and interstitial water of the sediments.

3. The concentrations of many interstitial nutrients are typically orders of magnitude higher than water column values (McRoy et al. 1972, Kenworthy et al. 1982, Short 1983a, Thayer et al. 1984), leading many

investigators to hypothesize that seagrasses derive most of their inorganic macronutrients from the sediment pore water (McRoy and Barsdate 1970, Iizumi et al. 1982, Short 1983b). Part of the reason these plant communities are so productive comes from the fact that they can obtain nutrients from the large sediment reservoir.

4. Although the sediments may be a large source of nutrients, several studies have suggested that seagrasses, and Z. marina in particular, can be nutrient limited. Microorganisms competing directly with the plants for inorganic nitrogen have been shown to assimilate as much as 49% of the regenerated ammonium in eelgrass bed sediments (Iizumi et al. 1982). Since ammonium is the dominant form of inorganic nitrogen in anaerobic sediments (Iizumi et al. 1980; Iizumi and Hattori 1982; Short 1983a,b), microbial assimilation may deprive the plants of nearly half the available nitrogen. Short (1983a) manipulated the density and biomass of Zostera in experimental plots and showed that sediment interstitial ammonium concentrations were inversely related to seagrass abundance. Since the kinetics of phosphorus and nitrogen uptake are concentration dependent (Penhale and Thayer 1980, Iizumi and Hattori 1982, Thursby and Harlin 1982, Short and McRoy 1984), nutrient depletion due to a large demand by the seagrasses eventually could result in nutrient limitation (Short 1983a,b). Modeling studies in conjunction with nitrogen uptake kinetics, carbon productivity, and nutrient regeneration rates suggest that nutrient limitation may occur under combinations of high plant density and low sediment organic matter (Short 1981, Short and McRoy 1984).

5. Under certain conditions seagrasses may respond to nutrient limitation by altering their morphology, density, and chemical composition (Bulthuis and Woelkerling 1981, Short 1983a, Short and McRoy 1984, Short et al. 1985). Orth (1977) added fertilizer containing nitrogen and phosphorus to surface sediments in an eelgrass bed. The plants increased in length, biomass, and total number of shoots compared to unfertilized control plots. This led Orth to conclude that eelgrass beds in Chesapeake Bay were nutrient limited. In a laboratory experiment, Roberts et al. (1984) demonstrated that sediment additions of balanced 14:14:14 (% nitrogen:phosphorus:potassium) and unbalanced (18:6:12) formulations of slow-release fertilizer stimulated growth of Zostera marina seedlings. Much less growth stimulation occurred when Zostera beds in Rhode Island were enriched with fertilizer that was released into the water column (Harlin and Thorne-Miller 1981). In relatively

quiescent areas, nutrient additions only stimulated macroalgal production, but where currents approached approximately 12 cm/sec, Zostera growth was enhanced.

6. Nutrient additions with slow-release fertilizers also have been applied to seagrass transplants, but with inconclusive results. Orth and Moore (1982a,b) reported that growth of transplanted plugs of Zostera marina could be stimulated by the initial, one-time addition of a balanced, slow-release fertilizer, but response varied dramatically between study sites. Since an increased rate of new shoot addition will increase survival potential and promote areal coverage by transplanted seagrasses, fertilization of these plantings may improve the probability of success. Any improvement in transplanting success should translate directly into enhancement of fishery habitat. Orth and Moore's results were untested with H. wrightii, however, and were based on spring plantings in a different geographic location using a plug technique that includes indigenous sediments and nutrients being transferred to the planting site. The method employed in this study (after Fonseca et al. 1982) uses sediment-free bare-root transplants. Further, in Orth and Moore (1982a,b), Roberts et al. (1984) and Pulich (1985), the nutrient release rates of the fertilizers were not reported, making it difficult to ascribe effects to a particular element. Thus, this study was for the purpose of further examining the use of fertilizers to enhance the population growth rate of Zostera and Halodule transplants in North Carolina. Reported herein are the results of field experiments examining the influence of slow-release fertilizers (in combinations of dosage and formulation) on survival, population growth, coverage rate, and sexual reproduction of Zostera marina and Halodule wrightii. This study also examined the productivity of selected transplants. This information, together with population growth data, was used to evaluate the cost versus benefit of subsidizing transplants with fertilizer.

PART II: METHODS

Site Selection

7. Three study sites were selected within Back Sound, Carteret County, N. C. (Figure 1). Sites were chosen based on their proximity to natural seagrass beds, and hence, their potential to uniformly support seagrass growth. The unvegetated areas that were chosen were free of active, anthropogenic impacts, such as dredging and filling, and human traffic. One site was a 2-year-old dredged material disposal site, while the other two were unvegetated spaces among existing meadows. A second site requirement was low sediment organic matter content (<2%). Kenworthy et al. (1982) demonstrated that on nearby shoals in Back Sound, unvegetated substrate with similar organic matter content had approximately one-half the exchangeable and dissolved NH_4 of vegetated substrate. Therefore, sites with low sediment organic matter content were selected to minimize the possibility of any nutrient loading from the sediment reservoir, and instead, to optimize fertilizer treatment effects.

Experimental Design

8. Three experimental plantings were performed between October 1984 and May 1985. The first experiment was a fall planting of Zostera (October 24), the second was a spring planting, also of Zostera (March 13), and the third was a late spring planting of Halodule (May 9). Each experimental planting (fall, spring, late spring) consisted of seven treatments replicated at each of the three study sites (Figure 2). There was no within-site replication in order to generalize at the regional rather than the site level. The seven treatments consisted of two different controlled-release fertilizers added in three dosage levels, plus one control (no fertilizer). The release rate was given by the manufacturer as 3 months at 70°F based on data in terrestrial systems. The treatment combinations were as follows:

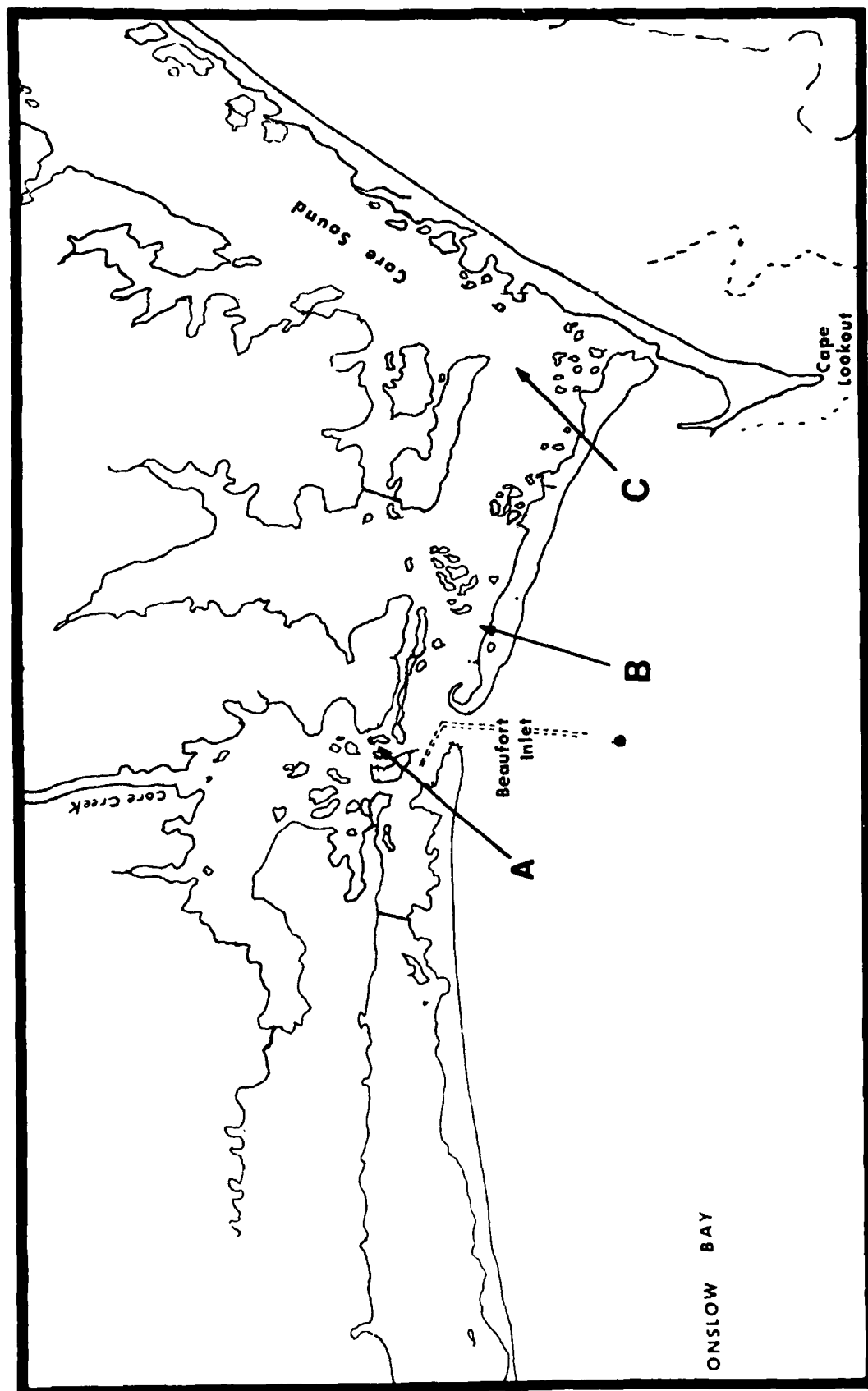


Figure 1. Location of the three planting sites: A = Kirby-Smith Island, B = Shackleford Shoal; C = Dredge Island

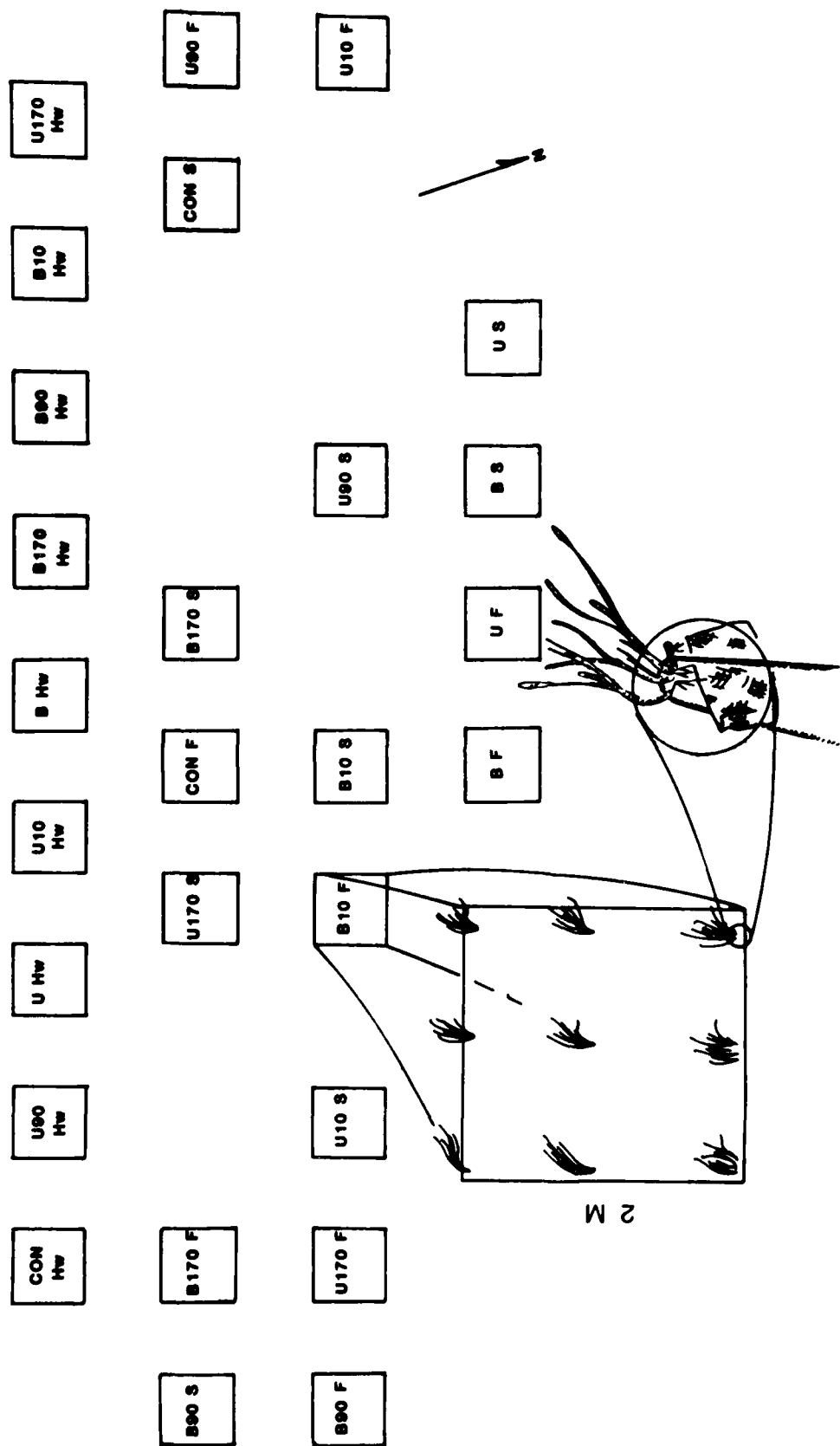


Figure 2. Layout of Dredge Island planting site as an example of treatment placement. Treatment labels represent balanced or unbalanced fertilizer, dose level in grams, and spring *Zostera* (S), fall *Zostera* (F), and *Halodule* (Hw) experiments. Treatments without dose levels are additional plots for periodic harvest of fertilizer samples. Blowup shows arrangement of planting units in a treatment and arrangement of anchoring pin and fertilizer bag

Treatment No.	Description
1	10 g balanced (14-14-14)*
2	10 g unbalanced (18-0-0)**
3	90 g balanced
4	90 g unbalanced
5	170 g balanced
6	170 g unbalanced
7	control (no fertilizer)

* Balanced ratio refers to the equal percentages of the total fertilizer weight reported by the manufacturer as nitrogen (6.6% NH_3 , 7.4% NO_3) - phosphorus (14% P_2O_5) - potassium (14% K_2O).

** Unbalanced ratio refers to the unequal percentages of the total fertilizer weight; according to the manufacturer, only nitrogen (8.5% NH_3 , 9.5% NO_3) was available.

9. Within each treatment were nine planting units (PU's), each assembled with 10 to 15 seagrass shoots attached to a 6-in. (15-cm) metal staple (as an anchor) with a 3-in (7.6-cm) twist tie (Fonseca et al. 1982, 1985) (Figure 2). Prewedged amounts (10, 90, 170 g) of pelletized fertilizer were placed in 1.1-mm-mesh plastic bags stapled closed and buried in the sediment immediately beneath each PU at the time of planting. Plastic mesh bags were used to enable periodic harvest of fertilizer for nutrient analysis. In order to apply the results of the nutrient analysis to the various treatments, it was felt that plastic mesh bags should be used consistently throughout the study. The staple of the PU was often secured through the plastic mesh bag. These nine PU's were planted on 1-m centers, creating 2- by 2-m plots for each treatment (Figure 2). Treatments were randomly assigned to locations on each study site for each experiment.

Environmental Conditions

Sediment characteristics

10. Surface sediment samples were taken at the time of planting from all treatments at each site to characterize the sediments and assess homogeneity within and between sites. Samples of the top centimeter of sediment were placed in an oven at 90° C and allowed to dry to a constant weight. After drying, each

sample was pulverized with a mortar and pestle to ensure that any particles consolidated by the drying process were disaggregated. Samples then were sieved in a sediment shaker for 20 min using standard sieve mesh sizes. Particle size distributions of each sample were characterized after Inman (1952), and phi mean, deviation, skewness, and kurtosis were calculated after Folk and Ward (1957). Two subsamples were taken from each sample before sieving for percent organic matter determination by combustion at 500° C for 12 hr.

Light, temperature, and salinity

11. Transmission of light through the water column was recorded at two randomly selected high and low tides at each site each month using a Sea Tech 25-cm transmissometer. Data were recorded as attenuation coefficients k for each site to compare the sites on a light energy basis. Average water depth (z) relative to mean sea level and a pooled yearly attenuation value (k) were used in the equation:

$$I(z) = I(o) e^{-kz}$$

where

$I(z)$ = light at depth z

$I(o)$ = incident light at sea surface

Given that $I(o)$ is the indicated photosynthetically active radiation (PAR), the value determined by e^{-kz} (a value between 0 and 1.0) is a factor by which incident light is reduced as a function of attenuation and depth.

12. Water temperature and salinity measurements were recorded from the mid-water depth on site using a glass thermometer and refractometer, respectively, at the same two randomly selected high and low tides as described above. Monthly averages of both factors were compiled for seasonal comparisons.

Current measurements

13. Water current velocity was measured at each location during a full moon tidal cycle to estimate regular, maximum velocities affecting the plantings. Two propeller-type flowmeters and an electromagnetic current meter were read simultaneously every hour during an entire tidal cycle. The maximum velocities over the tidal cycle were used to classify the hydrodynamic conditions of the sites (Fonseca et al. 1983).

Sediment flux rate

14. When the treatment locations were marked, one corner stake of each treatment was set so that the top of the stake was 50 cm above the sediment surface. At two randomly selected sampling times each month, the height of the top of this stake above the sediment was measured to assess sediment erosion or accretion. The absolute value of the fluctuation was divided by the number of days since the last measurement to yield the sediment flux rate. The average of these values was used to typify sedimentary dynamics at each site. The natural log (ln) of percent survival of PU's was regressed on the sediment flux rate to determine the potential influence of this factor on response of the plantings to fertilization.

Fertilizer Release

15. We suspected that the environmental conditions of this study were sufficiently different from the conditions under which the manufacturer determined the fertilizer release rates. Hence, we established an experiment to measure the release rates of the fertilizer during the study.

16. As each experiment (fall Zostera, spring Zostera, and late spring Halodule) was set up, two additional 2- by 2-m treatments were laid out and planted. Each of the nine PU's in each additional treatment was planted with a 90-g bag of fertilizer--one treatment having balanced and the other having unbalanced fertilizer types. Each time seagrass growth of that experiment was monitored (see paragraph 17), one randomly selected PU and fertilizer bag was recovered from each of these additional treatments at each site. The fertilizer was removed from each bag, rinsed in fresh water, freeze-dried, ground with a mortar and pestle, and stored in a vacuum dessicator until processed. The ground fertilizer was assayed for nitrogen (N) using a Carlo-Erba model 1106 elemental analyzer standardized with Acetanilide (percent N = 10.36). Phosphorus (P) was extracted by a potassium persulfate digestion; inorganic P was determined by a standard colorimetric method (Koroleff 1983). Phosphorus was extracted from the fertilizer at an efficiency approaching 100% of the manufacturer's specified level. Standard curves were prepared for inorganic nitrogen and phosphorus to cover the entire range of unknown values for the fertilizer.

Population Growth, Coverage, and Survival

17. Survival of PU's, the number of shoots, and the area covered per PU were recorded approximately every 5 to 7 weeks for each treatment in each experiment with time 0 counts obtained on the planting date. In each experiment at a given sample date, three randomly selected PU's per treatment (excluding additional plots) were censused for shoot abundance and areal coverage. Survival census included all PU's per treatment at that time. The total number of shoots in each randomly selected PU were counted by divers. The area covered was determined by placing a 0.25-m² quadrat subdivided into 5- by 5-cm squares over the PU. The number of sections completely filled by seagrass was recorded. Those squares on the edge of the PU that appeared to be half or less than one-half covered were recorded as half squares. All full and half squares were totaled to give the area covered by that PU at that time. During the May 1985 population census, the number of flowering shoots for each randomly selected Zostera PU was recorded.

18. The population and coverage data for the various treatments were compiled as regressions of ln-transformed population growth versus time since planting. These regressions were compared using an analysis of covariance (ANCOVA). Since there was no within-site replication, treatment replicates were composed of the three similar treatments from the three planting sites for each experiment. Comparisons using ANCOVA were made among the seven treatments within experiments (fall Zostera, spring Zostera, late spring Halodule) among balanced, unbalanced, and control groups within experiments, and between fall and spring plantings (Zostera) to determine if fertilizer type or dosage affected population growth and coverage rates. Density comparisons also were made between these fall and spring Zostera plantings. Extensive field notes and photographs, especially regarding faunal activities, were made by the divers during each site visit.

Net Shoot Productivity Measurements

19. Individual plants from the fall Zostera planting were tagged using adhesive aluminum tape (Kenworthy 1981). Ten shoots were tagged per treatment on April 5, 1985, at each of the three sites for a total of 30 shoots per treatment (210 total). At the time of tagging, the tip of the youngest (shortest) leaf was clipped in order to identify it later. The length and width of the clipped leaf and the next older leaf were measured. Weights of

these leaves were estimated using a regression of dry weight vs. surface area. In approximately 14 days, the entire plant was recovered. At that time, the lengths of the clipped leaf, next oldest leaf, and any new leaves were measured. The number of new shoots produced was observed as well. The amount of plant material produced since the tagging was determined by summing the weight of new leaf material produced since tagging. This was obtained by weighing the clipped leaf, the next oldest leaf and any new leaves and then subtracting the original estimated weight of the clipped leaf and the next oldest leaf. The plant material produced since the time of tagging was dried for 24 hr to a constant weight and recorded. The weight was divided by the number of days from tagging to harvest to give an estimate of production expressed as weight per shoot or grams dry weight per gram original weight per day. Comparison of production from the different treatments, replicated by site, gave an indication of any fertilizer effects by type and dose level.

Cost-Effectiveness of Fertilization

20. The cost of fertilization was estimated by timing divers in separate field trials engaged in transplanting Zostera with fertilizer packets in paper envelopes. The added time to produce the fertilizer packets and install the fertilizer packet in the sediment and the cost of the fertilizer were the only factors included in estimating cost. Benefit was determined by taking the number of shoots at a given time (e.g., time 0, day 50, 100, 150, 200, and 250) as determined by the population growth regression line and dividing the cost per shoot at time 0 by that number. The treatment(s) that had the greatest reduction in cost per shoot, especially in comparison to the unfertilized control treatments, was considered to be the most cost-effective. Any increase in net shoot production was not directly included in an estimate of benefit.

PART III: RESULTS

Sediment Particle Size and Organic Content

21. Particle size analysis revealed a very uniform sediment type within and between sites. The mean and median particle size for all three sites were all equal and ranged between 0.17 and 0.34 mm. All particle size distributions were classified as moderately sorted, nearly symmetrical or slightly negatively skewed, and mesokurtic or leptokurtic after the definitions of Folk and Ward (1957).

22. The percent organic matter of the sediment at all sites ranged from 0.35 to 1.03% of the sediment dry weight. Only the Kirby-Smith Isl. site had a value above 1.0%; the rest were never higher than 0.65% of the sediment dry weight. These values met our criteria for planting in relatively low organic content sediments.

Water Depth

23. Water depths varied considerably between sites while within-site variation was minimal. Low-tide water depths at the Shackleford Shoal site ranged between 36 and 42 cm (mean = 39 cm) while high-tide depths were more consistent and averaged approximately 95 cm. The Dredge Isl. sites were slightly deeper with a low-tide depth range of 36 to 70 cm (mean = 56 cm) while high-tide depth averaged 99 cm. Kirby-Smith Isl. was similar in low-tide depth to the Dredge Isl. site with a range of 49 to 62 cm (mean = 56 cm) but with a high-tide average of 131 cm.

Light Regime

24. The light regime of the three sites varied between high and low tides, seasons, and within the sites. High tides had appreciably better water column transmissivity than low tides with attenuation coefficients often being lower by 1.0 at high tide (Table 1). There also was a consistent increase in overall water column transmissivity from fall to late spring (Table 1). Differences between sites were greatest at the low-tide samples. The Dredge Isl. site tended to be clearer, followed by the Shackleford Shoal site, leaving the Kirby-Smith Isl. site as the most turbid. These attenuation coefficients (k) may be related to Secchi disk k_{SD} values by the following empirically derived equation:

$$k_{SD} = 0.0615 k + 0.365 \quad (r = 0.785)$$

using an equation for k_{SD} developed by Weinberg (1976).

Table 1. Average transmissometer attenuation coefficients (k) by site, experiment, and tide for transplants of Zostera marina and Halodule wrightii

<u>Experiment</u>		<u>Low Tide</u>	<u>High Tide</u>
<u>Shackleford Shoal</u>			
<u>Zostera</u>	(Fall)	3.65	2.74
<u>Zostera</u>	(Spring)	3.34	2.02
<u>Halodule</u>	(Late spring)	2.96	1.72
<u>Dredge Island</u>			
<u>Zostera</u>	(Fall)	3.61	2.89
<u>Zostera</u>	(Spring)	2.39	2.13
<u>Halodule</u>	(Late spring)	2.15	1.88
<u>Kirby-Smith Island</u>			
<u>Zostera</u>	(Fall)	4.85	2.65
<u>Zostera</u>	(Spring)	4.59	2.65
<u>Halodule</u>	(Late spring)	3.86	2.48

25. These k values and water depth measurements were used to compute the relative percent incident light energy (%Iz) that should have been reaching the plants in the various treatments at each site. It was then possible to regress population growth rates of the individual treatments for each experiment (fall Zostera, spring Zostera, late spring Halodule) on %Iz to determine if any differences in response to fertilization instead could be attributed to different light regimes between treatments. Percent incident light ranged from 12 to 19 in the fall experiment. When population growth rates for the fall experiment treatments were regressed on %Iz for that time, only 11% of the variation in population growth rates could be accounted for by a negative relation with %Iz. In the spring planting, 25% of the variation in population growth could be accounted for by %Iz, but again this relation was negative with a range in %Iz from 20 to 25 across treatments. The late spring Halodule planting demonstrated a positive relation between growth and %Iz, but the relation could only account for 7% of the variation in growth as a function of light. Percent incident light ranged from 26 to 35 in this experiment.

Temperature and Salinity

26. Temperature ranges during the study were within normal limits for this area (Figure 3) (Fonseca et al. 1985). Temperature tended to be lower on incoming tides. The range for all three sites was between a low of 6° C in February and a high of 30.5° C in July (Figure 3).

27. Salinity showed a steady increase during the course of the study. Starting with a low of 28 to 30 ppt in October, salinity increased steadily to May and June, peaking at 35 to 37 ppt (Figure 3). Again, high tides tended to be slightly more saline than low or ebbing tides. By July, however, there was evidence of decreasing salinity.

Current Regime

28. Currents were all within the low-velocity range (< 50 cm/sec) as defined by Fonseca et al. (1983). Shackleford Shoal had a peak flow of 40 cm/sec; the Dredge Isl. 13.5; and Kirby-Smith Isl. 27.8. The velocity at the Shackleford Shoal site was high enough to move the sediment on a regular basis. Of the three sites, the Dredge Isl. site had the greatest exposure to wind-generated waves with a northeasterly fetch of approximately 13 km before encountering the subtidal sandbag dike surrounding the area. This dike

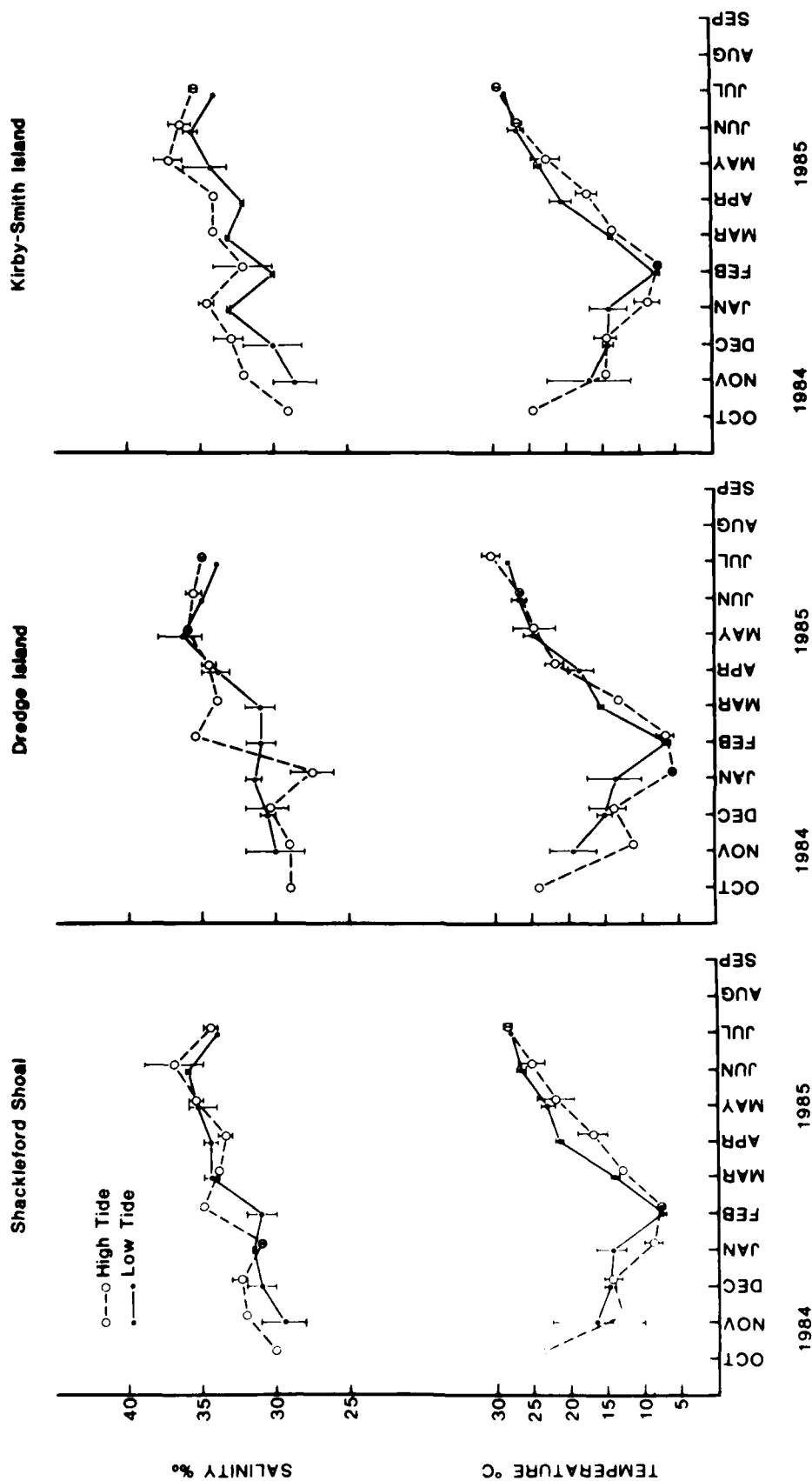


Figure 3. High and low tide salinity and temperature for the three planting sites over the course of the study

eliminated most wave energy. Shackleford Shoal was moderately exposed from the northwest with a fetch of approximately 3 km. Kirby-Smith Isl. was the most protected, with the longest fetch being no more than 300 m.

Sediment Flux Rate

29. Only in the late spring experiment (Halodule) did sediment flux rate (SFR) have a correlation coefficient >0.5 with PU survival. Approximately 2 to 10% of the variation in PU survival could be attributed to sediment flux in the fall and spring Zostera experiments. In the late spring experiment, a transformation applied to the percent survival data demonstrated that 69% of the variation in PU survival could be attributed to the SFR ($\ln \% \text{ PU survival} = 43.14 (\text{SFR}) - 140.8, r^2 = 0.69$). It was during the Halodule experiment that PU loss occurred.

Fertilizer Release

30. Nitrogen loss from the fertilizers during the fall experiment was linear over 286 days. The balanced (14-14-14) fertilizer released approximately 0.044% of the original amount per day (Figure 4). The unbalanced fertilizer released 0.058% of the original amount per day (Figure 4). For the balanced fertilizer treatments, this release rate meant that 0.0043, 0.0385, and 0.0729 g N were released per fertilizer bag per day for the 10-, 90-, and 170-g dose level treatments, respectively. Unbalanced treatments at the same dose levels released 0.0052, 0.0473, and 0.0894 g N PU⁻¹ day⁻¹.

31. The spring experiment suffered from an apparent failure by a new batch of unbalanced fertilizer to release any nitrogen (Figure 5). The balanced fertilizer released nitrogen at a rate of 0.068% of the original fertilizer weight per day, yielding 0.0068, 0.0611, and 0.1155 g N PU⁻¹ day⁻¹ for the 10-, 90-, and 170-g dose level treatments, respectively. This rate was higher than the fall and was probably due to the warmer temperatures between March and July as opposed to the October to July growth time of the fall experiment (Figure 5).

32. The late spring experiment, which used only Halodule, had the highest nitrogen release rates and occurred during the warmest time of the year (Figure 5). Balanced fertilizer nitrogen loss was 0.088% of the original fertilizer weight per day (0.0088, 0.0796, and 0.1503 g N PU⁻¹ day⁻¹ for each dose level) while unbalanced release was 0.113% per day (0.0113, 0.1014, and 0.1915 g N PU⁻¹ day⁻¹ for each dose level).

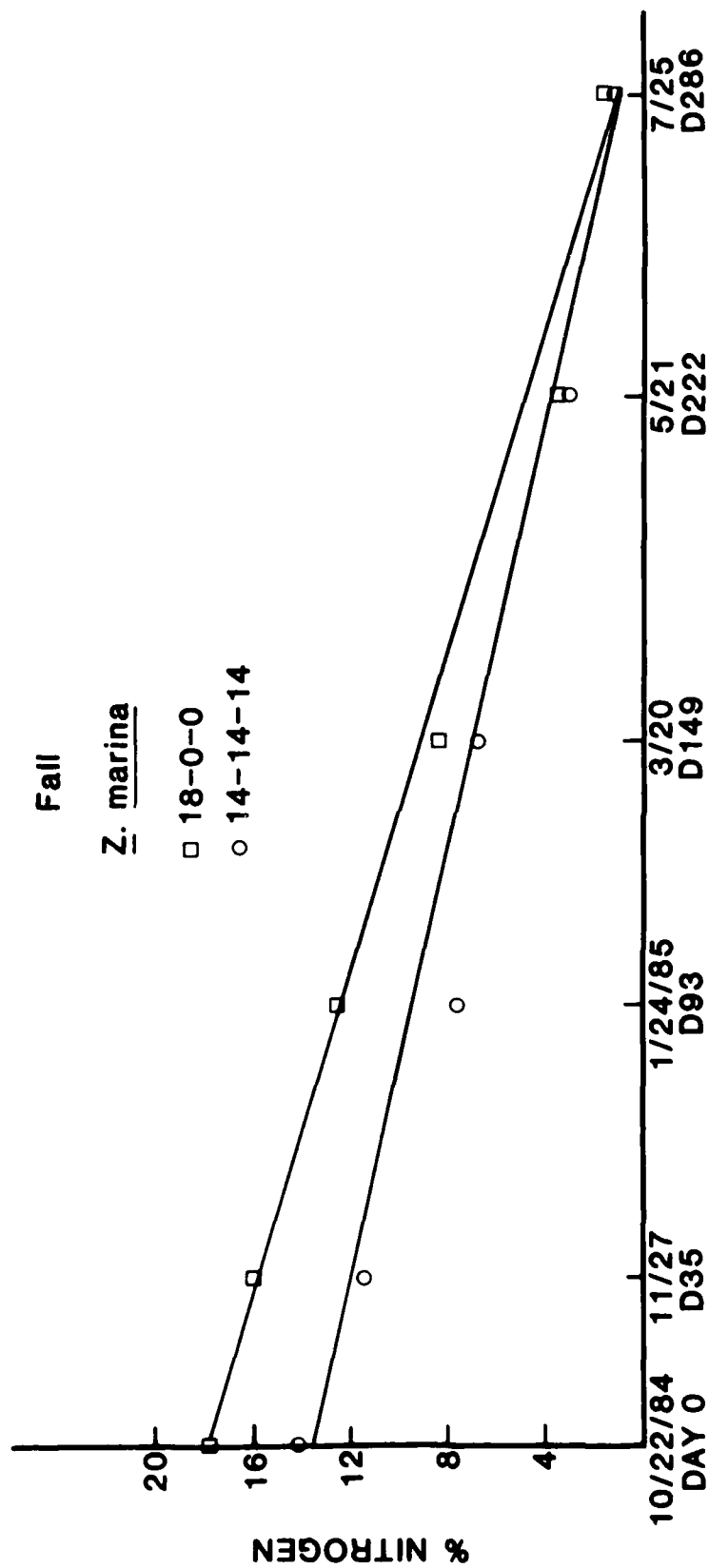


Figure 4. Average percent nitrogen content of fertilizer (unbalanced: 18-0-0 and balanced: 14-14-14) over the course of the fall Zostera planting

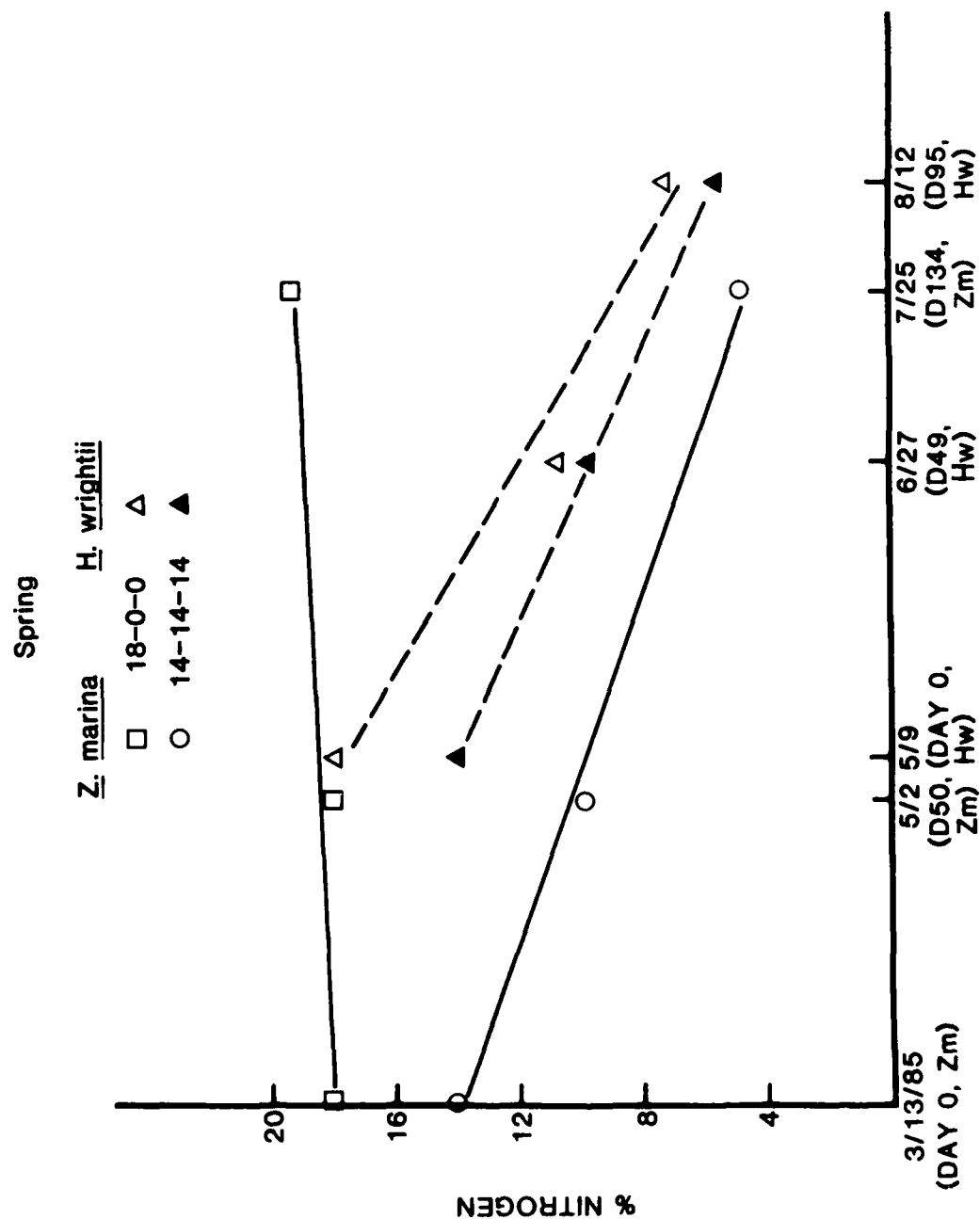


Figure 5. Average percent nitrogen content of fertilizer (unbalanced: 18-0-0 and balanced: 14-14-14) over the course of the spring Zostera and Halodule plantings

33. Contrary to the performance of nitrogen, there was no net release of phosphorus in any of the balanced treatments (Figures 6 and 7). During the fall Zostera planting, fertilizer phosphorus content began to decline after 150 days but was still higher than the initial values at 286 days.

Planting Unit Survival

34. By day 243 in the fall Zostera experiment, survival of PU's varied tremendously. The range of survival was from 11 to 100%. The Kirby-Smith Isl. site had the lowest overall survival at this time, but as in all treatments at all sites, there was no pattern of loss that could be correlated with fertilizer treatments (Table 2). Planting unit survival in the spring Zostera experiment was higher (Table 2), but this experiment ran only 134 days. Survival of the late spring Halodule planting was poor (Table 2). The Shackleford Shoal Halodule planting site did not survive to day 59 while the Kirby-Smith site lost between 60-90% of its PU's. The Dredge Isl. site fared better, with PU's present in all treatments at day 93 (Table 2).

35. After the summer of 1985, no further data were recorded on PU growth. Surveys performed in fall 1985 indicated that few of the experimental treatments had survived. At the Shackleford Shoal site, no Halodule survived and only a few PU's from the spring Zostera experiment remained. Survival of the fall Zostera experiment was much the same as reported for day 243 (Table 2). At the Dredge Isl. site, only five of the Halodule treatments still had PU's that appeared capable of surviving to the next growing season. No other experimental treatments survived at this site. After the passage of Hurricane Gloria, no PU's survived at the Kirby-Smith Isl. site due apparently to sediment movement.

Population Dynamics

Shoot addition

36. Data on shoot numbers were ln-transformed and regressed on time. These regressions were plotted and compared initially by treatment within experiments using ANCOVA. A significant difference ($p < 0.05$) was found between the slopes of the regression lines only for treatments in the fall Zostera experiment. All treatments with balanced fertilizer had regressions with numerically higher slopes; this was evident also from the regression plots (Figure 8). Treatments were then grouped by fertilizer type (balanced, unbalanced, control) without regard to dose level (10, 90, or 170 g/PU) and compared as before using ANCOVA.

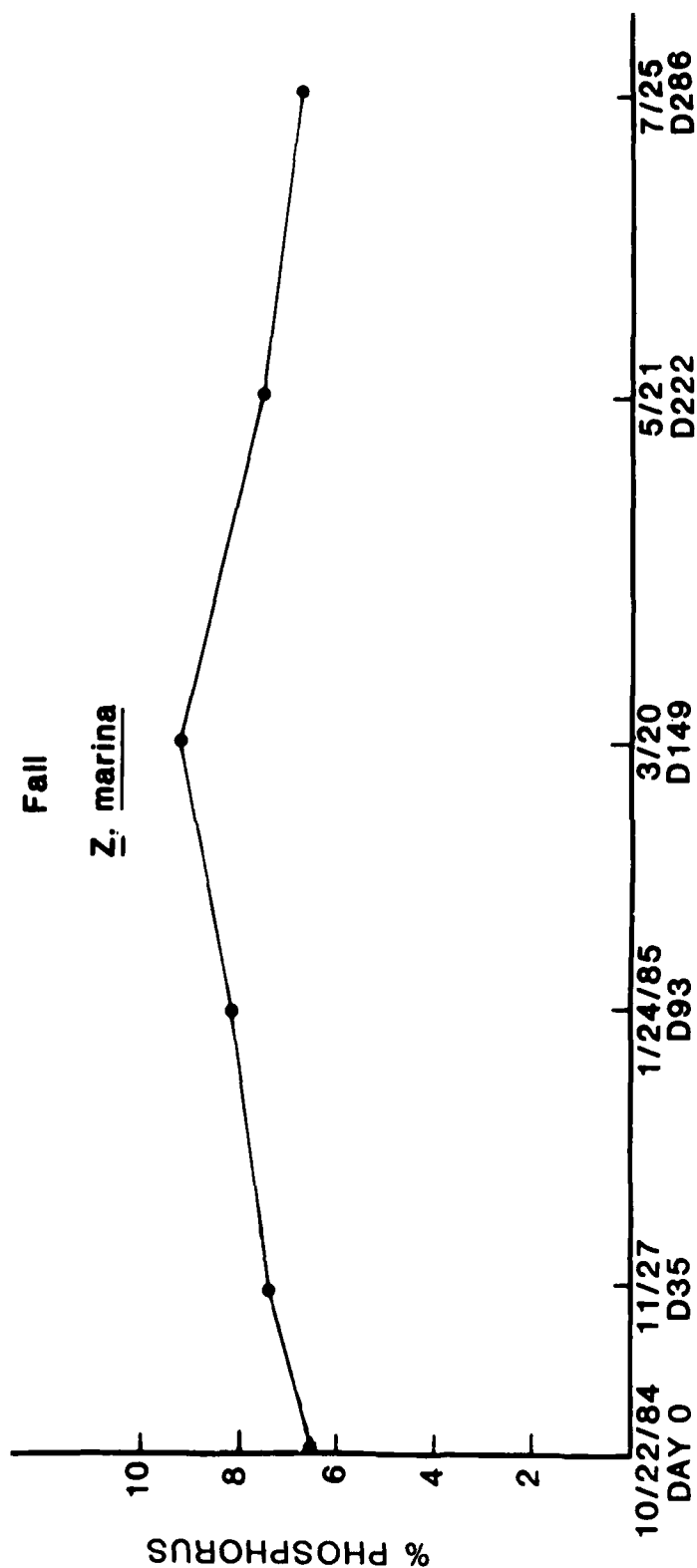


Figure 6. Average percent phosphorus content of fertilizer (balanced only 14-14-14) over the course of the fall Zostera planting

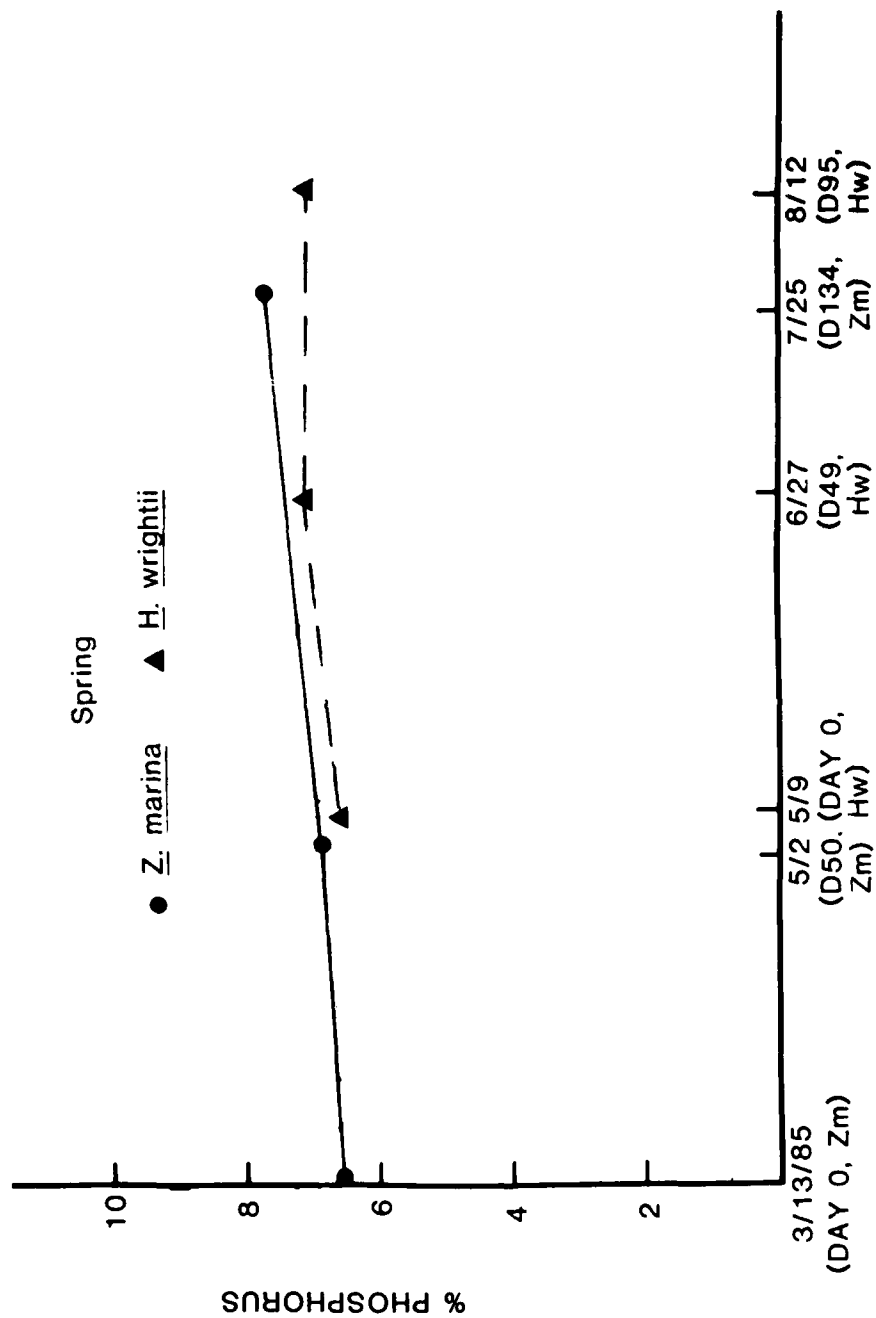


Figure 7. Average percent phosphorus content of fertilizer (balanced only 14-14-14) over the course of the spring Zostera and Halodule plantings

Table 2. Percent planting unit survival by treatment, experiment, and site for transplants of Zostera marina and Halodule wrightii.

	<u>Treatment</u>						
<u>Experiment</u>	<u>B10</u>	<u>U10</u>	<u>B90</u>	<u>U90</u>	<u>B170</u>	<u>U170</u>	<u>Control</u>
<u>Shackleford Shoal</u>							
Fall <u>Zostera</u>							
Day 59	88.9	100	66.7	88.9	33.3	100	100
Day 243	77.8	100	66.7	77.8	33.3	100	100
Spring <u>Zostera</u>							
Day 56	66.7	100	55.6	100	66.7	44.4	100
Day 125	66.7	100	55.6	100	66.7	44.4	100
Spring <u>Halodule</u>							
Day 59	0	0	0	0	0	0	0
Day 93	0	0	0	0	0	0	0
<u>Dredged Material Island</u>							
Fall <u>Zostera</u>							
Day 59	100	100	77.8	100	100	77.8	100
Day 243	100	88.9	77.8	44.4	88.9	11.1	77.8
Spring <u>Zostera</u>							
Day 56	100	100	100	88.9	100	88.9	77.8
Day 125	100	100	100	88.9	100	88.9	77.8
Spring <u>Halodule</u>							
Day 59	11.1	66.7	55.6	100	55.6	44.4	44.4
Day 93	11.1	66.7	55.6	100	44.4	44.4	44.4
<u>Kirby-Smith Island</u>							
Fall <u>Zostera</u>							
Day 59	66.7	88.9	100	100	77.8	77.8	100
Day 243	55.6	33.3	55.6	77.8	77.8	55.6	33.3

(Continued)

Table 2 (Concluded)

	<u>Treatment</u>						
<u>Experiment</u>	<u>B10</u>	<u>U10</u>	<u>B90</u>	<u>U90</u>	<u>B170</u>	<u>U170</u>	<u>Control</u>
<u>Kirby-Smith Island (contd)</u>							
Spring <u>Zostera</u>							
Day 56	77.8	100	33.3	66.7	33.3	100	66.7
Day 125	66.7	100	33.3	66.7	33.3	100	66.7
Spring <u>Halodule</u>							
Day 59	0	44.4	0	44.4	11.1	11.1	55.6
Day 93	0	44.4	0	44.4	11.1	11.1	55.6

Notes: B10, 90, and 170 = balanced (14-14-14 nitrogen, phosphorus, and potassium) fertilizer at dose level in grams. U10, 90, and 170 = unbalanced (18-0-0) fertilizer at dose level in grams. Control = unfertilized plantings.

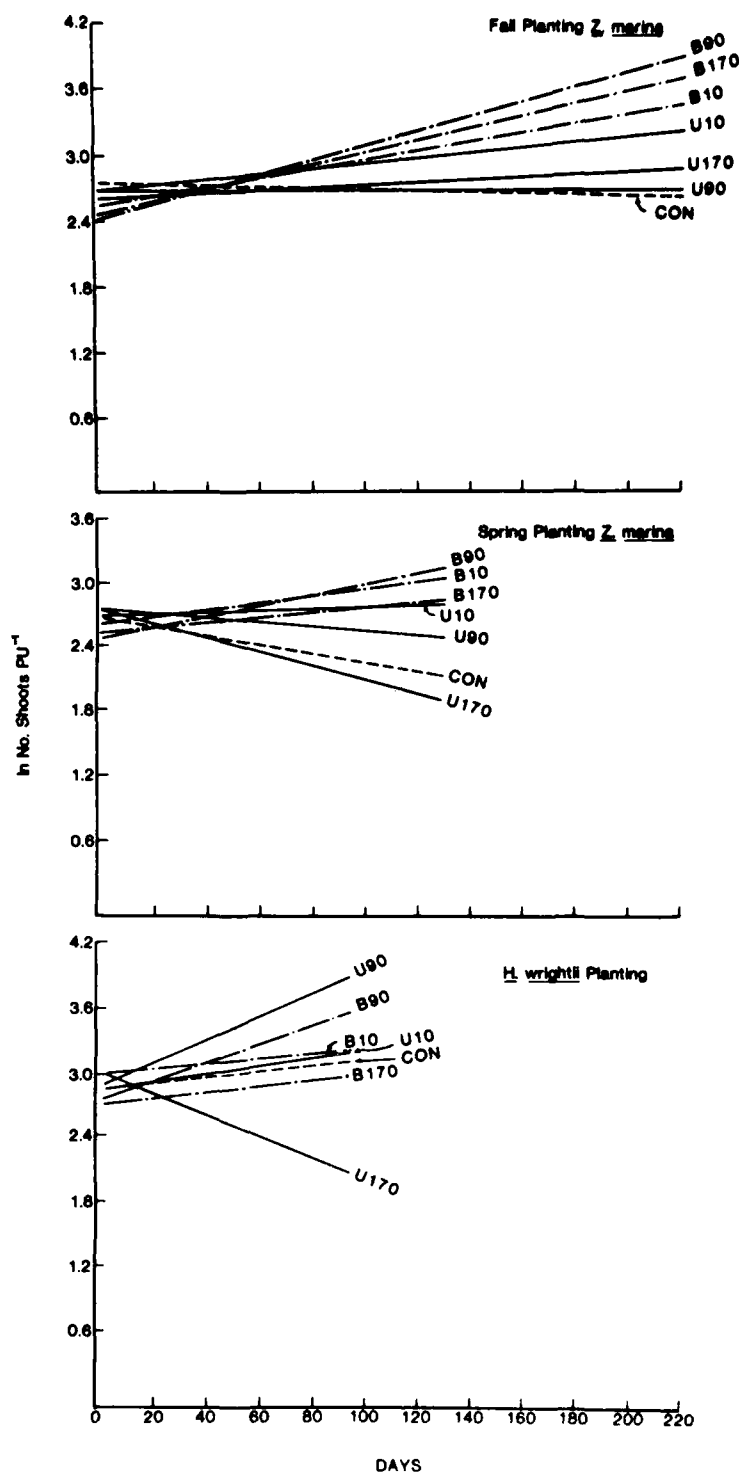


Figure 8. Natural log-transformed plots of number of shoots per PU by treatment and experiment over time. Treatments B10, B90, and B170 = balanced fertilizer (14-14-14) and dose level in grams; U10, U90, and U170 = unbalanced (18-0-0) fertilizer and dose level in grams; con = control or unfertilized planting

A significant difference ($p < 0.05$) was found between the three dose levels in the fall Zostera experiment. The graphical representation of these groups (Figure 9) suggested that the balanced treatments had a greater slope than either unbalanced or control treatments, which appeared to be identical. Thus, the balanced group was eliminated to compare the unbalanced and control groups by ANCOVA. No significant difference ($p < 0.05$) was found between the slopes or adjusted group means of the unbalanced or control treatments. It was concluded that balanced fertilizer additions in the fall Zostera experiment produced a significantly greater rate of shoot addition per PU over time. Further analysis of similarly grouped treatments for the spring Zostera and late spring Halodule experiments produced no significant differences.

37. Comparisons were next made of the fall versus spring experiments for Zostera. The fall-to-spring comparisons were made among the balanced, unbalanced, and control groups of the two experiments. No significant differences were found in the three comparisons.

38. The numerical abundance of flowering shoots was not significantly different between spring and fall plantings (one-way analysis of variance, ANOVA, $\alpha < 0.10$, data not shown). The percent flowering shoots per PU was, in all cases except the unbalanced 10-g dose level, higher in the spring Zostera planting (Figure 10). ANOVA (one-way) was used to compare control versus balanced and unbalanced Zostera treatments within and between both seasons. There were no significant differences ($\alpha < 0.10$) within seasons. Significant differences ($\alpha < 0.10$) were found between seasonal comparisons (fall and spring) of balanced, unbalanced, and control treatments grouped irrespective of dose level. No flowering was observed in the Halodule plantings.

Areal coverage

39. Comparisons were made of areal data as for shoot data (Figure 11). There were no significant differences between the slopes of the regression lines in any of the three experiments. There were significant differences ($p < 0.05$) in the adjusted group means between balanced 10-g and the control and between balanced 170-g treatments and the control in the fall experiment. This suggested that while the rates of increase in area did not differ between treatments, the actual area of a PU at any time would be greater in the balanced 10- and 170-g treatments than the control in the fall experiment.

40. Data also were grouped by fertilizer type, irrespective of dose level, as was done with the shoot data and compared using ANCOVA (Figure 12). Using

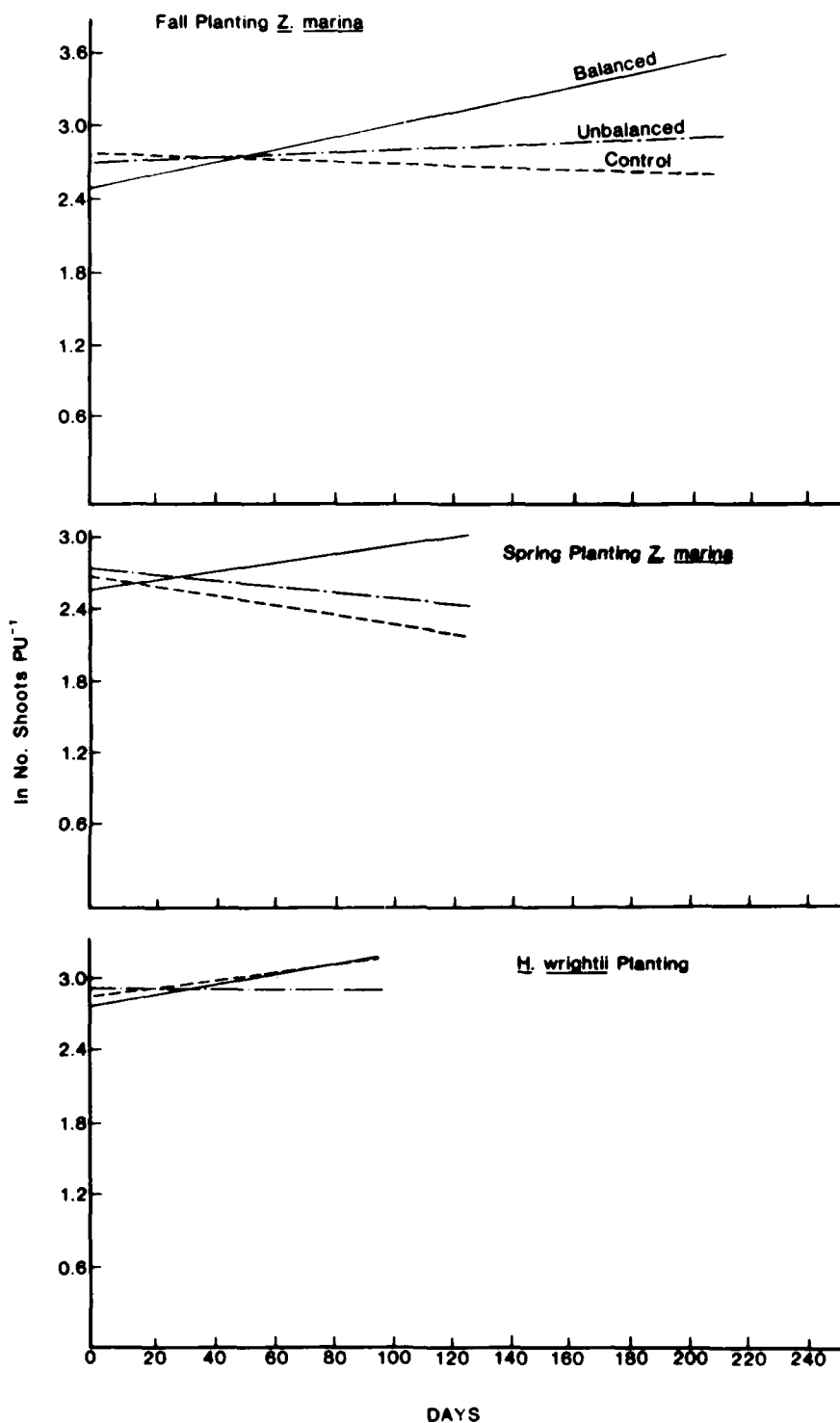


Figure 9. Natural log-transformed plots of number of shoots per PU by treatment and experiment over time. Balanced (14-14-14) and unbalanced (18-0-0) treatments refer to all dose levels of a given fertilizer type pooled together, whereas control refers to all unfertilized plantings pooled together

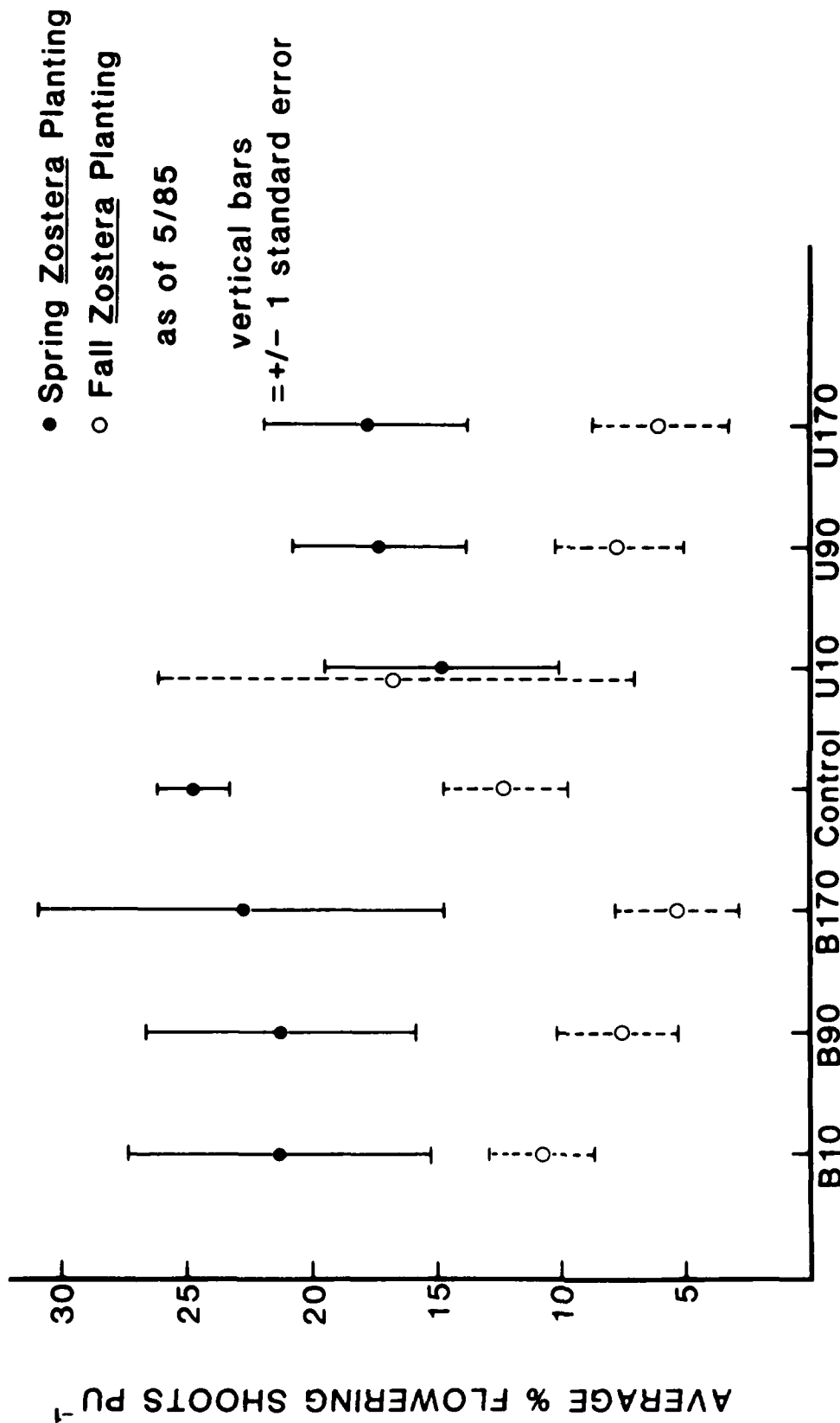


Figure 10. Average percent flowering shoots per PU for fall and spring Zostera plantings by treatment. B10, B90, and B170 = balanced (14-14-14) fertilizer at each dose level in grams; U10, U90 and U170 = unbalanced (18-0-0) fertilizer at each dose level in grams; Control = unfertilized plantings

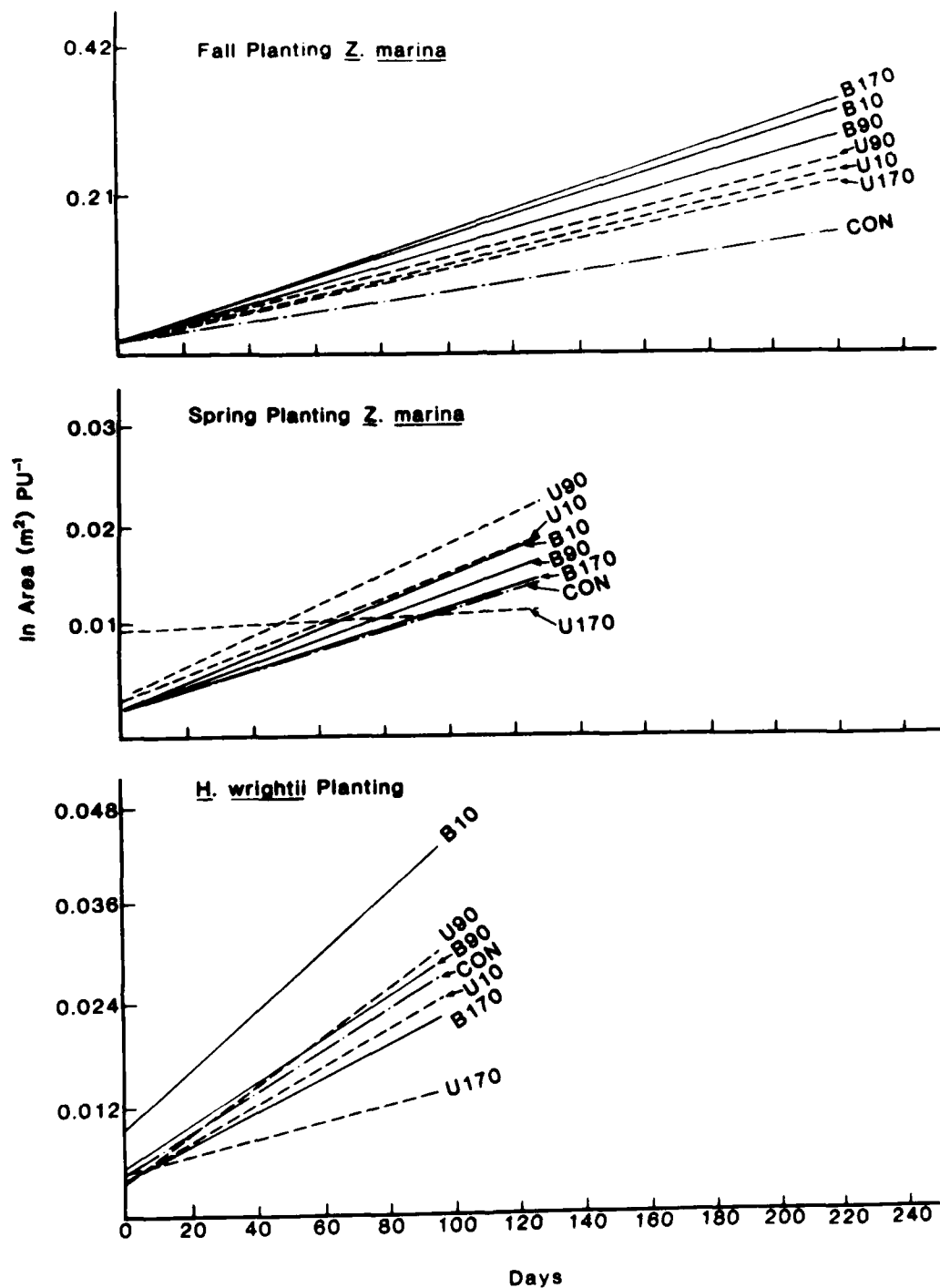


Figure 11. Natural log-transformed plots of area (m^2) covered per planting unit by treatment and experiment over time. Treatments B10, B90, and B170 = balanced fertilizer (14-14-14) and dose level in grams; U10, U90, and U170 = unbalanced (18-0-0) fertilizer and dose level in grams; Con = control or unfertilized planting

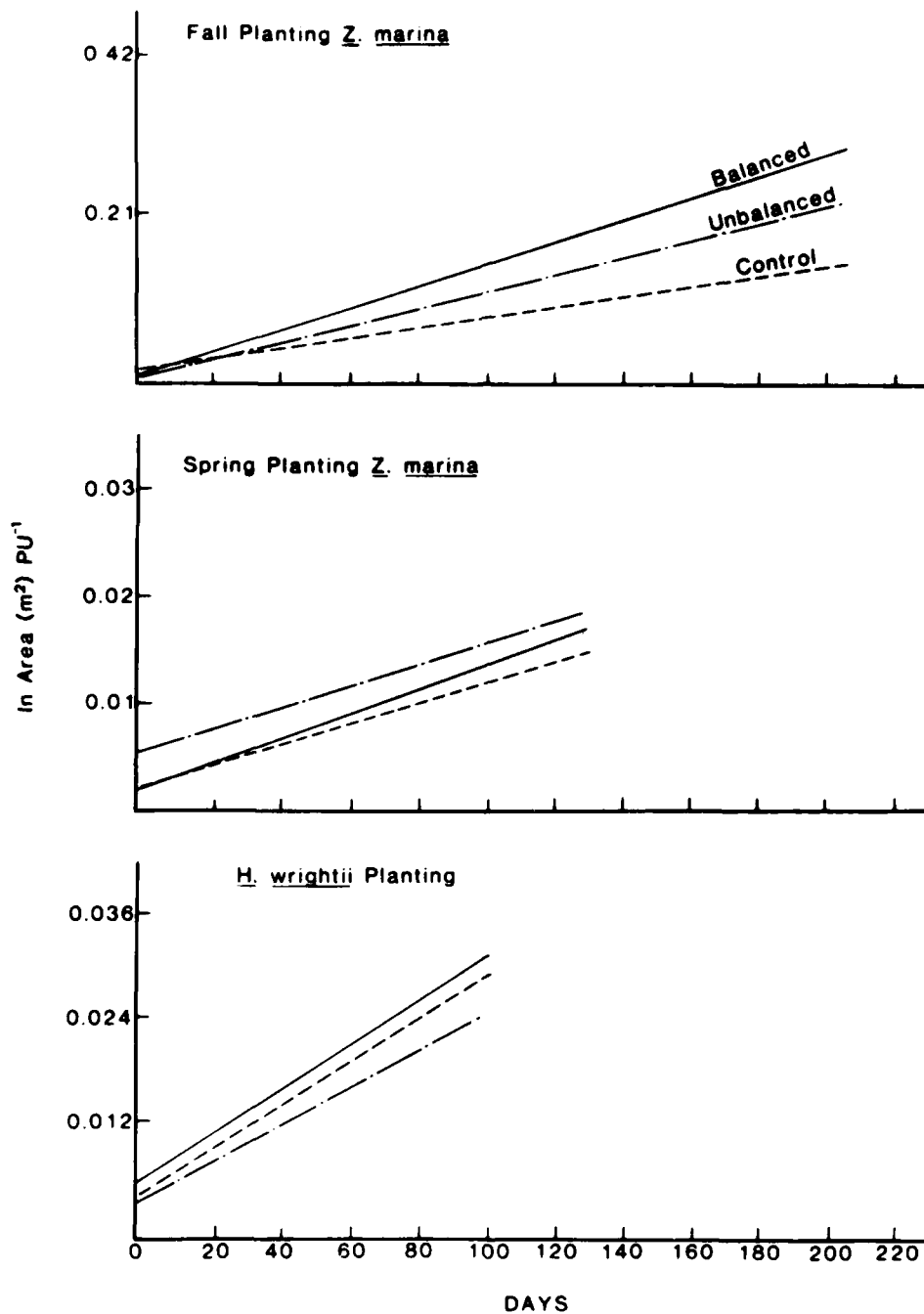


Figure 12. Natural log-transformed plots of area (m²) covered per PU by treatment and experiment over time. Balanced and unbalanced treatments refer to all dose levels of a given fertilizer type pooled together whereas control refers to all unfertilized plantings pooled together

ln-transformations of the area data produced no significant differences in regression line slopes or adjusted group means among any of the three groups (balanced, unbalanced, or control).

41. The grouped ln-transformed data also were compared between fall and spring experiments (both Zostera), but no significant differences were found. There was no difference in slopes or adjusted group means between control plots of the fall and spring experiments.

Shoot density

42. There were dramatic differences in the density of shoots between experiments, but not between treatments. The fall Zostera experiment had densities of $< 300/\text{m}^2$ while the spring Zostera and late spring Halodule had densities $> 1,000$ and often $> 2,000$ shoots/ m^2 . The high shoot density was to be expected for Halodule, which is typically found in very dense assemblages such as this, but was not expected for Zostera.

Productivity

43. Of the 30 shoots that were tagged for production in each fall Zostera treatment (10/treatment/three sites), between 18 and 23 were recovered. The average balanced treatment productivity regardless of dose level was 0.0058 g/shoot/day, with a range of 0.005 to 0.0065 (Figure 13). The average, unbalanced treatment productivity was 0.003 g/shoot/day with a range of 0.001 to 0.004. Control treatments averaged 0.003 g/shoot/day. No productivity comparisons were made with the spring Zostera or Halodule plantings.

Cost-Effectiveness of Fertilization

44. The reduction in cost per shoot for the fall Zostera experimental treatments is given in Table 3. The balanced treatments with dose levels of 10 and 90 g/PU had a cost per shoot equal to or less than that of the control plantings by the end of the 250-day growing season. The higher population growth rates of these treatments not only reduced cost per shoot to the level of control (unfertilized) treatments, but the area covered by these treatments was significantly higher (Figures 11 and 12). The balanced 170 g/PU dose level did not reduce cost per shoot as effectively. In the unbalanced treatments, only the 10 g/PU dose level reduced costs comparably to those of the balanced treatments (Table 3). The 90 and 170 g/PU dose level treatments had costs per shoot four to six times greater than those of the other treatments. No cost per shoot estimates were made for the spring Zostera or Halodule plantings.

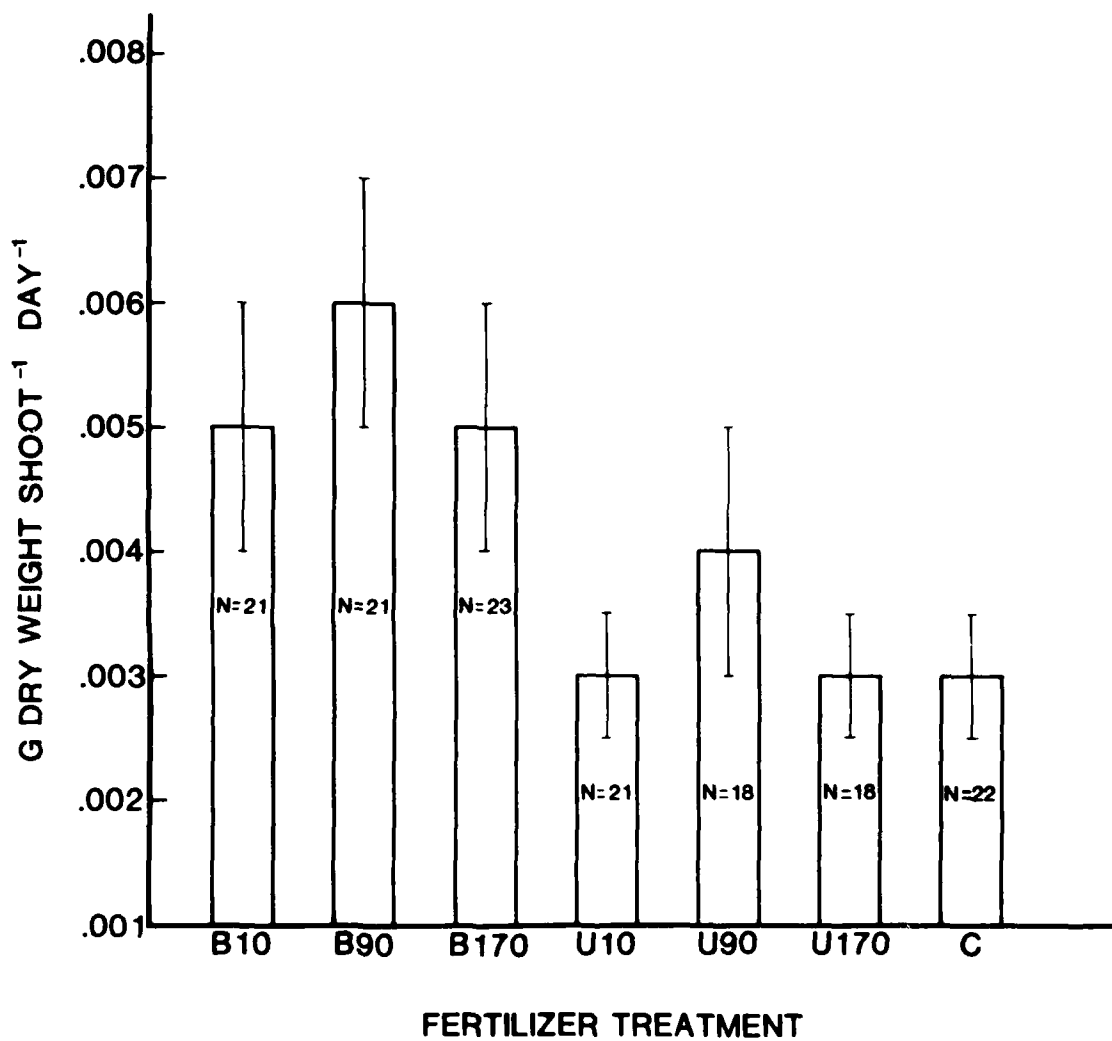


Figure 13. Productivity (grams dry weight/shoot/day) of fall *Zostera* plantings by treatment. B10, B90, and B170 = balanced (14-14-14) fertilizer at each dose level in grams

Table 3. Cost per shoot of fall-initiated transplants of Zostera marina over time as a function of fertilizer treatment

Treatment	Time 0 No. Shoots = 16					
	T0	T34	T50	T150	T200	T250
B10	0.012	0.016	0.012	0.008	0.007	0.005
B90	0.023	0.031	0.023	0.012	0.008	0.006
B170	0.035	0.054	0.035	0.020	0.015	0.011
U10	0.012	0.011	0.011	0.009	0.008	0.007
U190	0.023	0.021	0.025	0.025	0.025	0.025
U170	0.035	0.048	0.037	0.033	0.031	0.029
C	0.007	0.007	0.007	0.006	0.006	0.006

Notes: B10, 90, and 170 = balanced (14-14-14) fertilizer at dose level in grams. U10, 90, and 170 = unbalanced (18-0-0) fertilizer at dose level in grams. Control (C) = unfertilized plantings.

PART IV: DISCUSSION

45. Throughout the study period, consideration was given to the possible influence of local environmental conditions on seagrass growth. Variations in growth as a result of systematic differences in environmental conditions between sites and treatments had the potential to cause an erroneous interpretation of fertilizer effects on the transplants. However, no systematic pattern of growth response to environmental conditions that would alter the study conclusions on fertilizer effects could be detected.

46. Temperature and salinity were within normal limits for this area (Thayer et al. 1984, Fonseca et al. 1985), and adjacent, natural beds appeared healthy and vigorous.

47. The low organic matter content and relative lack of fine particle sizes within our sites indicated that low-nutrient reservoirs existed (Kenworthy et al. 1982, Fonseca et al. 1983). Low-sediment nutrient reservoirs meant that the level of exchangeable nutrients would also be low, enhancing the potential for influence on seagrass growth by the fertilizer additions during this study.

48. Differences in available light as a function of depth and local turbidity were considered to have the greatest potential for confounding fertilizer effects. Correlations were found between population growth rates and light regimes (paragraph 25). The relation between growth and light for the fall and spring Zostera plantings was considered spurious since the relation was negative; i.e., growth decreased with increased light. The range of light levels for these treatments (12 to 25% of ambient) did not appear to be high for limitation photoinhibition. The Halodule planting had a positive correlation of growth with light, but the relation could account for only 7% of the variation in the population growth. It was concluded that variation in light regimes did not contribute to the pattern of growth responses observed between sites.

49. The only environmental factor that had a selective effect among sites was the sediment flux rate. This factor affected the percent survival of the PU's. Although the three sites are considered low-current regimes (after Fonseca et al. 1983), that classification does not account for wind-driven circulation. A storm event occurred shortly after planting of the Halodule experiment, resulting in a rapid loss of a significant number of plantings (Table 2). While other plantings lost a greater percentage of their numbers over time, only the Halodule plantings could be correlated with sediment fluctuation (paragraph 29).

50. One of the objectives of this investigation was to determine if fertilization could significantly improve initial survival of plantings. After eliminating the Halodule treatments described above because of their relation to sediment fluctuation, the percent survival results were reviewed (Table 2). However, no pattern of survival could be conclusively linked to fertilizer treatment. It was concluded that the fertilizer additions used in this study did not affect initial survival of PU's.

51. Consideration was then given to the effects of the fertilizer on population growth, flowering, area covered, and productivity by first examining the nature of the fertilizer release. Except for the unbalanced spring Zostera experiment which did not exhibit any nutrient release, enough nitrogen was released even by the 10-g dose levels to meet the requirements of the seagrass at all times. Although one cannot be sure that the nitrogen reached all shoots equally, given the point source application and the radial growth of the seagrass away from that point, enough was released to meet daily requirements. This was determined by multiplying the number of shoots at any given time by production values (Kenworthy, unpublished) to estimate g C/planting unit/day. Using a C:N tissue ratio of 16:1 (Thayer et al. 1984), the number of grams N needed/PU/day was estimated. This number was always exceeded by the release rates (see paragraphs 30-32). However, the absence of phosphorus release from the balanced fertilizer prevents the conclusion that the significantly higher growth and coverage ratio in the fall Z. marina experiment were due to the additive effects of nitrogen and phosphorus release.

52. The ranges of nitrogen release were not substantially different between balanced and unbalanced treatments to explain the difference (paragraphs 30-32) and, as stated above, even the lowest dose level (balanced, 10-g) released enough nitrogen per day to meet plant needs. According to the manufacturer's specifications, nitrogen should release faster than phosphorus in the balanced 14-14-14 formulation. This case shows an extreme difference in release rates, where nitrogen released at an adequate rate although, up to day 149, the percentage of phosphorus in the fertilizer appeared to increase (Figures 6 and 7). This may not be a true increase in phosphorus, but rather as nitrogen and possibly potassium released, phosphorus became a larger proportion of the total weight remaining. On the other hand, some phosphorus also may have been adsorbed or exchanged on the resin coating with the phosphorus in the sedimentary environment, leading to a net increase in fertilizer phosphorus

content. After day 149 there was a net decrease in the phosphorus content of the fertilizer, yet the phosphorus remaining at the end of the experiment (day 286) still exceeded the initial phosphorus value.

53. According to the manufacturer, this fertilizer is used primarily in well-aerated, well-drained soils and there is no technical information available to compare with the release characteristics observed in the natural, anaerobic marine sediments. Unfortunately, the three published experimental studies utilizing this fertilizer with seagrass did not report the release characteristics (Orth and Moore, 1982 a,b; Roberts et al. 1984). All the authors have assumed a relatively constant release rate for all elements in the fertilizer over a 3-month period (see Roberts et al. 1984, p 322). It is impossible to determine whether phosphorus was actually released from the fertilizer in these previous studies, but our experience suggests that it was not.

54. It is possible, however, to evaluate the possible impact of a manufacturer's quality control problem with regard to the resin coating. As described earlier, the unbalanced 18-0-0 was a custom-blended experimental fertilizer which, since initiation of this study, the manufacturer has ceased producing. Difficulties were encountered with coating the ammonium sulfate particles so that complete coverage of the particles could not be obtained consistently without overcoating the fertilizer. It is likely that the failure of N to release from the 18-0-0 in the spring Zostera planting was due to overcoating. Although there was concern that differential coating may have been responsible for lack of phosphorus release in the balanced formulation, the quality control problem was ruled out as in this case since nitrogen was released from the same 14-14-14 application. It is suspected that the P_2O_5 (phosphoric acid) and the resin coating exhibit characteristics that are substantially different in the physicochemical regime of the anaerobic, marine sediments than under the conditions with which the manufacturer tested the fertilizer. Had previous investigations examined the release characteristics of the fertilizer rather than simply assuming they would not vary, it would be much easier to evaluate the use of those fertilizer products and to determine the interactions of nitrogen and phosphorus in affecting the growth and coverage by these seagrasses.

55. It is similarly difficult to explain Zostera density, flowering, and productivity as a function of fertilizer treatment. Differences in density

and percent flowering may be attributed solely to the time of planting. There was no significant difference (one-way ANOVA, $\alpha = 0.10$) in percent flowering between treatments within each of the spring and fall Zostera experiments. There was a significant difference among balanced, unbalanced, and control treatments between the spring and fall Zostera treatments (one-way ANOVA, $\alpha = 0.10$). The percent flowering was higher in the spring Zostera planting, possibly because there had not been sufficient time since planting for a large number of new, vegetative shoots to be added. In contrast, the fall Zostera planting had 5 months more to increase the number of new vegetative shoots. The percent of the original number of shoots per PU that flowered (also expressed as the absolute number per PU) was the same between spring and fall, suggesting that induction of flowering occurred late in 1985. Although these data do not indicate any fertilizer effect on flowering, they do support our previous contentions that Zostera should be planted in the fall in the middle Atlantic region to optimize vegetative shoot production (Fonseca et al. 1982, 1985).

56. The disparity in densities between the spring and fall Zostera transplant also may be explained simply as an artifact of planting time. The fall planting gave adequate time for shoot numbers to increase until the January-February slowdown in growth typical for this area (Kenworthy et al. 1982). Planting in March corresponded with a vigorous spring initiation of growth with many of the original shoots beginning to flower. There was little spreading from those shoots, while the rest appeared to branch prolifically. By fall 1986, these new shoots probably would have spread out, and normal densities would have been found. In any event, these densities do not appear to be specifically related to fertilizer additions.

57. Productivity measurements of the fall Zostera plantings were significantly greater in the balanced versus unbalanced and control treatments. Given the overlapping ranges of N-availability and lack of phosphorus release, we cannot provide a convincing explanation of why balanced treatments had a higher level of productivity than unbalanced treatments. The only other difference between the fertilizer treatments was the presence of potassium in the balanced additions. The fertilizer samples were not tested for potassium release, mainly because there was no basis for assuming that this element was limiting. If consistent increases in productivity could be attained from fertilizer addition, the addition of the added plant material to the detrital

food chain should be factored into the cost-effectiveness of the fertilizer additions.

58. There are several alternative explanations that need to be evaluated in future investigations to resolve the unexplained, apparent effect of the balanced fertilizer on seagrass population growth and productivity. This may simply be an artifact of inadequate replication even though the experimental design is valid. It is known that significant differences in transplanted seagrass growth occur between sites, seasons, and years (Fonseca et al. 1982, 1985). The experiments could simply have produced a fortuitous pattern of response that erroneously suggests that balanced fertilizer substantially improves growth. The positive results of Orth (1977), Orth and Moore (1982a,b), Roberts et al. (1984) with similar fertilizer, however, make acceptance of these trends very tempting. However, as described earlier, the performance of fertilizer in those studies is unknown. Also, it should be noted that Orth and Moore (1982a) performed transplants at numerous locations in Chesapeake Bay and found positive results at only one of those sites (Orth and Moore 1982a,b). Such a pattern of response supports the "fortuitous pattern" alternative suggested above. Another explanation could be that different physiological states of seagrass between spring and fall, and perhaps between site and year, have prevented us from observing a consistent pattern of response to fertilizer between these different times. Clearly, our knowledge of the nutrient requirements of these seagrasses and the chemistry of fertilizer release is so incomplete as to warrant further investigation before reliable conclusions can be drawn.

59. Without an examination of the fertilizer release as done here, the cost-effectiveness of balanced fertilizer in fall Zostera plantings could be accepted. The increased level of shoot addition offset the increased cost per shoot generated by the one-time addition of fertilizer. Fertilizer costs were amortized in one growing season simply by the higher shoot generation rate. Even if these results could be accepted without reservation, given what we know about the fertilizer release, there is another danger in interpreting these data. If one plots the number of shoots per planting over time for the fall Zostera balanced, unbalanced, and control experiments against the previous 4-year average growth for the Beaufort area (Fonseca et al. 1985) a surprising trend is evident (Figure 14). The best population growth observed in this study was correlated with balanced fertilizer addition, but was no greater

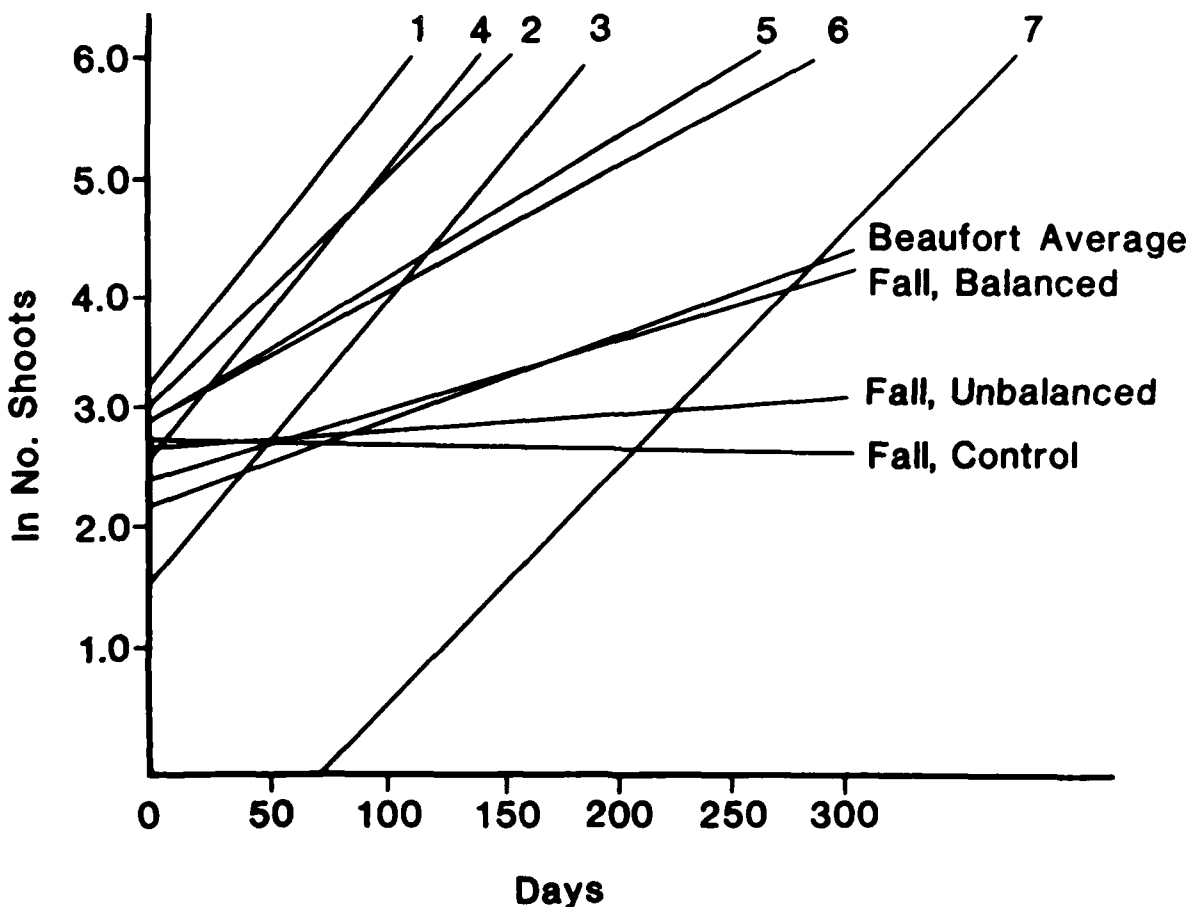


Figure 14. Population growth of *Zostera marina* in: 1) a natural undisturbed population in Denmark (Verhagen and Nienhuis, 1983); 2,3) annual populations in Rhode Island (Harlin et al. 1982); 4,5) fertilized, transplanted plugs of *Zostera marina* in the lower Chesapeake Bay, Virginia (Orth and Moore, 1982,a,b); 6) unfertilized, transplanted plugs of *Zostera marina* in lower Chesapeake Bay, Virginia (Orth and Moore, 1982 a,b); 7) seedling of *Zostera marina* recolonizing a disturbance site in North Carolina; Beaufort average = average value for eight transplant experiments in North Carolina; fall balanced = average value for all transplants treated with balanced (14-14-14) fertilizer; fall unbalanced = average value for all transplants treated with unbalanced (18-0-0) fertilizer; fall, control = average value for all transplants not treated with fertilizer

than the average growth for the area over the last few years. What this demonstrates is that year-to-year (and probably site-to-site) variation in growth can negate any anticipated gains from fertilizer additions. This is true even if we accepted fertilizer effects without reservation. The danger lies in these fertilizer addition data being interpreted as allowing a transplanter to reduce the number of plantings (density of PU) at time 0 in hopes of reducing cost. The strategy would be to let the fertilizer addition promote greater growth and thus attain a similar coverage but at a lesser overall cost.

60. Figure 14 demonstrates that if one had reduced PU numbers to less than the recommended number (Fonseca et al. 1982, 1985), even additional growth from fertilizer subsidies would have generated substantially less cover at the end of the year than expected. The performance of balanced fertilizer is even less impressive when compared with other areas and studies of both subsidized and unsubsidized populations (Figure 14). Although, because of latitudinal differences in growth, such a comparison might be unfair. These data do point out the danger in applying these results, even had they not been in question because of the fertilizer performance, to populations in other geographic locations.

61. It was concluded that despite the consistently higher population growth, coverage, and productivity generated by balanced fertilizer additions, this difference cannot be ascribed solely to the fertilizer. The lack of fertilizer release in a manner that could support these differences leads to the speculation that other factors, including other sediment chemical conditions, and contributing to the observed response of seagrass growth. The fertilizer release problem also casts doubts on the validity of previous studies that report enhanced growth from similar fertilizer additions. If an acceptable explanation could be developed that reconciles the fertilizer release performance with the observed seagrass response, it could be concluded that fertilizer addition may be cost-effective. Even so, annual variation in growth has demonstrated the potential to negate expected gains in coverage. Until further research is conducted that specifically investigates the role of identified chemical additions on the growth of seagrass, no alterations in recommended planting spacing should be considered. Despite our extensive cautioning, we must note that the fertilizer additions did significantly increase population growth, coverage, and productivity in a cost-effective

manner. The use of balanced fertilizer additions (10 g/PU) apparently has the potential to improve Zostera transplanting over unfertilized transplants, but we cannot make a conclusive statement about the reliability of the technique. Management and restoration plans should not consider fertilization as anything but a pilot experiment until the chemical cause and effect and additional replicated plots are established and analyzed.

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